Safety and Immunologic Activity of Multiple Infusions of
APN401 Wake Forest Baptist Comprehensive Cancer Center
CCCWFU 03716
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STUDY SYNOPSIS

IND AGENT	• APN401 is a suspension of autologous peripheral blood mononuclear cells (PBMCs) transfected with siRNA that reduces Cbl-b.
INDICATION	• Advanced solid tumors
PRIMARY OBJECTIVES	• Determine toxicities • Determine immunologic effects
SECONDARY OBJECTIVE	• Document clinical response and survival
STUDY PHASE	• Phase I
STUDY DESIGN	• Open-label
MAIN INCLUSION CRITERIA	• Signed written informed consent • Histologically confirmed metastatic or inoperable solid tumor no longer responding to standard therapies • Must be at least 4 weeks since prior systemic therapy, radiation and/or surgery • ECOG performance status 0-1 • Hematocrit ≥28%, WBC >3000/µl, and platelets >100,000/µl • Creatinine ≤ 1.6 mg/dl • ALT and AST<2.5 fold the upper limit of normal and total bilirubin ≤ 1.6 mg/dl
MAIN EXCLUSION CRITERIA	• Pregnant or breastfeeding patients • Untreated, progressing, or symptomatic brain metastasis • HIV, hepatitis B, or hepatitis C (active) infection • Active autoimmune disease • Ongoing requirement for an immunosuppressive treatment • Significant cardiovascular or pulmonary disease
TREATMENT PLAN	• PBMC for infusions will be collected by leukapheresis the day prior to infusion. • Patients will be administered three infusions separated by 4 weeks • Blood will be drawn pre-therapy and at 3, 5, 9, and 10 weeks, and then every 8 weeks for immune assessments • Imaging will be performed pre-therapy and 2 weeks after infusion 3 and then every 8 weeks for clinical response • Six patients will be treated initially. If a dose limiting toxicity (DLT) is observed in ≤ 1/6, than an additional four patients will be enrolled to allow evaluation of laboratory correlates. Enrollment will be suspended if >1/6 or >3/12 patients manifest DLT
LABORATORY CORRELATES	• PBMC cytokine response • Circulating immunocytes
POPULATION SIZE	• Two to 12 eligible and evaluable patients
STUDY DURATION	• Patients will be followed for toxicity until objective disease progression or 1 year, whichever occurs first.

Contents

1.0 Background

1.1 Adoptive cellular immunotherapy

One approach of addressing immune deficiencies preventing an antitumor response in vivo effectively applied to improve survival is to infuse immune cells activated ex vivo. Sipuleucel-T, an autologous cellular immunotherapy, has been approved for the treatment of metastatic prostate cancer [1]. Sipuleucel-T is manufactured from peripheral blood mononuclear cells (PBMCs) isolated during leukapheresis that are cultured ex vivo with PA2024, a fusion protein consisting of prostatic acid phosphatase and granulocyte macrophage colony stimulating factor, and then infused back into the patient. Other strategies of activating, expanding, and/or engineering immune cells ex vivo to mediate antitumor effects have exhibited considerable promise in a variety of malignancies [2,3]. Cellular immunotherapy has not been reproducibly effective, and several obstacles remain, including sustaining the in vivo survival and function of transferred cells and generating high-avidity tumor-reactive T cells. T-cell persistence can be transiently maintained by administering interleukin (IL-) 2, but this is often associated with significant toxicity and may also promote expansion of regulatory T (Treg) cells that limit antitumor immunity [4]. The poor T-cell function often observed has been more difficult to address, as it, in part, reflects deficient expression of costimulatory proteins and/or increased expression of inhibitory ligands or suppressive cytokines by tumor cells [5]. Thus, strategies to improve T cell survival, function, and avidity are needed.

Cell processing is performed per FDA approved SOPs. They include several tests of sterility.

1.2 Cbl-b

Cbl-b (Casitas B-lineage lymphoma b), an E3 ubiquitin ligase, is a major negative regulator of lymphocyte function. It increases the threshold for naive T-cell activation by regulating T-cell receptor and CD28 costimulatory signaling [6]. Abrogating Cbl-b expression rescues IL-2 production and proliferation by naive $CD28^{-/-}$ T cells. Following target recognition, interferon (IFN-) γ production is enhanced, and target avidity is increased. Importantly, neither proliferation or cytokine production is induced in unstimulated T cells, indicating that Cbl-b silencing enhances T cell activities only in the context of antigen stimulation and does not lead to an unwanted general activation of all lymphocytes. Tumors lack the costimulatory proteins that bind CD28, and CD28 expression is often lost on T cells that have differentiated to effector cells during repetitive stimulations [7]. Thus, disrupting Cbl-b expression in tumor-reactive T cells may provide a unique strategy for increasing autocrine IL-2 production for promoting proliferative responses and survival following tumor recognition in the absence of costimulation and may also reduce the threshold for T cell activation to allow low-avidity T cells to respond to tumors expressing limiting amounts of antigen. Cbl-$b^{-/-}$ knock-out mice are resistant to outgrowth of transplantable and spontaneous tumors [8,9]. Adoptive transfer of Cbl-$b^{-/-}$ $CD8^{+}$ T cells into tumor-bearing wild-type mice resulted in detectable therapeutic activity [9-11]. Tumor resistance has been attributed to the increased responsiveness of the small number of naive tumor-reactive Cbl-$b^{-/-}$ $CD8^{+}$ T cells present in the polyclonal repertoire. Genetic deletion of Cbl-b or targeted inactivation of its E3 ligase activity also have been shown to license natural killer (NK) cells to spontaneously reject metastatic tumors in mouse models [12]. Cellular immunotherapy with Cbl-$b^{-/-}$ cells or inactivation of Cbl-b E3 ligase activity does not result in

significant inflammation or in autoimmunity. Thus, abrogating Cbl-b expression in effector immune cells may improve the efficacy of adoptive cellular immunotherapy of human malignancies without significant clinical toxicity.

1.3 APN401

APN401 is a suspension of viable autologous human PBMCs that have been transiently transfected with siRNA in order to reduce Cbl-b protein levels, for autologous cellular immunotherapy applications. A highly efficient transfection protocol using a commercial electroporation device is used to simultaneously transfect T, B, NK cells and monocytes with the siRNA with minimal cell damage. Using this protocol shut down of Cbl-b expression can be achieved for more than seven days in stimulated human T cells, resulting in strong enhancement of T-cell activation, cytokine production, and proliferation. Cbl-b-silenced PBMCs display a clear increase in proliferation and IL-2 and IFN-γ production in response to anti-CD3/28 stimulation. In addition, Cbl-b silencing also increases secretion of tumor necrosis factor (TNF) α and IL-17. Adoptive transfer of Cbl-b silenced murine CD8 T cells using a similar transient siRNA silencing procedure as for human cells significantly decreased tumor burden [13]. Moreover, T cells isolated from tumor-draining lymph nodes displayed sustained activated T cell function five days after Cbl-b silencing and transfer. Little toxicity was observed. No unwanted effects of autoimmunity in other tissues were observed. Thus, APN401 is a novel approach to enhance antitumor immune responses. Moreover, because of potential of limited toxicity and synergy with other cancer therapies that have a mechanistic component of immune activation, APN401 has high potential to be included in combination programs.

1.4 Phase 1 study of single infusion of APN401

The feasibility and safety of a single intravenous infusion of APN401 were evaluated in patients with refractory metastatic solid tumors [14]. Patients with active autoimmune disease or a requirement for immune suppressive drugs were excluded. PBMC were collected by leukapheresis. The following day PBMCs were transfected with Cbl-b siRNA by electroporation to produce APN401 and then infused intravenously over 30 minutes. Three to ten patients were treated in each of the three dosing cohorts in which they received a single intravenous infusion of 5, 10, or 50 $x10^5$ cells/kg APN401. PBMC were successfully collected and transfected with Cbl-b siRNA in all patients enrolled. CD3 T cell, CD56 NK cell, CD19 B cell, and CD14 monocytes were transfected with similar efficiencies. Transfected PBMCs produced 4-fold more IFN-γ 2-fold more IL-2 in response to CD3/CD28 stimulation than PBMC that were processed and electroporated without siRNA. From 27.0 to 467 X 10^6 siRNA transfected PBMCs were infused. Infusions were well tolerated. Mild to moderate chills developed in most patients at the completion of the infusion. Dose-limiting toxicities were not observed. There was no immediate hypersensitivity and no evidence of autoimmune adverse effects. Stable disease observed in two patients with pancreatic cancer and one with colorectal cancer was associated with increases in PBMC responses to stimulation with CD3/CD8 and tumor-associated peptides.

1.5 Study rationale

Adoptive cellular immunotherapy with ex vivo manipulated autologous immune cells has been an attractive approach to refractory cancers. Cbl-b is a major negative regulator of immune function. Adoptive cellular immunotherapy with Cbl-b silenced cells significantly

decreased tumor burden in mouse models without toxicity. Multiple infusions were applied in these studies. APN401 is a suspension of autologous human PBMCs transfected ex vivo with a siRNA that silences Cbl-b developed for adoptive cellular immunotherapy applications. A phase I clinical trial to determine the toxicities of a single intravenous administration of APN401 established feasibility and safety. This clinical trial will examine multiple infusions and also provide an initial estimate of immunologic activity. The overall goal is to determine if there is sufficient evidence of safety and immunologic activity to proceed to phase II clinical trials, alone or in combination with other antitumor agents.

2.0 Objectives

2.1 Primary

2.1.1 To determine the toxicities and establish the safety of multiple infusions of APN401

2.2 Secondary

2.2.1 To determine the immunologic effects of multiple infusions of APN401

2.2.2 To document clinical response and survival

3.0 Selection of Patients

3.1 Inclusion criteria

3.1.1 Patients with histologically confirmed inoperable, recurrent or metastatic malignant solid tumors, deemed incurable, and who have either:

- failed to respond to standard therapy or
- for whom no standard therapy is available or
- refuse to receive standard therapies

The study is intended to enroll patients with pancreatic and colorectal cancer. Patients with other types of solid tumors will require approval by the Principal Investigator.

3.1.2 Measurable disease as defined by RECIST (see Section 9.0)

3.1.3 Patients with treated, stable, and asymptomatic brain metastases are eligible

3.1.4 Patients on every 2, 3 or 4 week systemic therapy programs must be off the treatment program for at least 2, 3, or 4 weeks, respectively, and must have recovered from any clinically significant toxicity experienced. Patients on weekly or daily systemic therapy programs and patients receiving radiation must be at least 1 week since treatment and recovered from any clinically significant toxicity experienced. Must be at least 4 weeks and have recovered from major surgery

3.1.5 Age ≥ 18 years

3.1.6 ECOG performance status 0 or 1

3.1.7 Patients must have the following lab values obtained < 4 weeks prior to starting treatment:
- WBC ≥ 3000/μL
- Platelets ≥ 100,000/μL
- Hematocrit ≥ 28%
- Creatinine ≤ 1.6 mg/dL
- AST and ALT < 2.5 x upper limit of normal
- Bilirubin ≤ 1.6 mg/dL (except patients with Gilbert's Syndrome, who must have a total bilirubin less than 3.0 mg/dL)
- Albumin ≥ 3.0 g/dL
- INR ≤ 1.5

3.2 Exclusion criteria

3.2.1 Women must not be pregnant or breastfeeding due to unknown effects of the study treatments on the unborn fetus. All women of childbearing potential must have a blood test within 72 hours to rule out pregnancy. Women of childbearing potential and sexually active males must be strongly advised to use an accepted and effective method of contraception. Women of childbearing potential (WOCBP) must be using an adequate method of contraception to avoid pregnancy throughout the study and for 26 weeks after the last dose of investigational product, in such a manner that the risk of pregnancy is minimized. Sexually mature females who have not undergone a hysterectomy or who have not been postmenopausal naturally for at least 24 consecutive months (i.e., who have had menses at some time in the preceding 24 consecutive months) are considered to be of childbearing potential. Women who are using oral contraceptives, other hormonal contraceptives (vaginal products, skin patches, or implanted or injectable products), or mechanical products such as an intrauterine device or barrier methods (diaphragm, condoms, spermicides) to prevent pregnancy, or are practicing abstinence or where their partner is sterile (e.g., vasectomy) should be considered to be of childbearing potential.

3.2.2 Untreated, progressing, or symptomatic brain metastases

3.2.3 Autoimmune disease, as follows: Patients with a history of inflammatory bowel disease are excluded as are patients with a history of symptomatic disease (e.g., rheumatoid arthritis, systemic progressive sclerosis [scleroderma], systemic lupus erythematosus, autoimmune vasculitis [e.g., Wegener's granulomatosis]). Patients with motor neuropathy considered of autoimmune origin (e.g., Guillain-Barre Syndrome and myasthenia gravis) are excluded. Patients with a history of autoimmune thyroiditis are eligible if their current thyroid disorder is treated and stable with replacement or other medical therapy.

3.2.4 Any other malignancy from which the patient has been disease-free for less than 2 years, with the exception of adequately treated and cured basal or squamous cell skin cancer, superficial bladder cancer or carcinoma *in situ* of the cervix.

3.2.5 Other ongoing systemic therapy for cancer, including any other experimental treatment. These include concomitant therapy with any of the following: IL-2, interferon, ipilimumab, pembrolizumab, nivolumab, or other immunotherapy; cytotoxic chemotherapy; and targeted therapies.

3.2.6 Ongoing requirement for an immunosuppressive treatment, including the use of glucocorticoids or cyclosporine, or with a history of chronic use of any such medication within the last 4 weeks before enrolment. Patients are excluded if they have any concurrent medical condition that requires the use of systemic steroids (the use of inhaled or topical steroids is permitted).

3.2.7 Infection with HIV. Due to the mechanism of action of APN401, activity and side effects in an immune compromised patient are unknown.

3.2.8 Active infection with Hepatitis B. Active or chronic infection with Hepatitis C.

3.2.9 Clinically significant pulmonary dysfunction, as determined by medical history and physical examination. Patients with a history of pulmonary dysfunction must have pulmonary function tests with a $FEV_1 \geq 60\%$ of predicted and a DLCO ≥ 55% (corrected for hemoglobin)

3.2.10 Clinically significant cardiovascular abnormalities (e.g., congestive heart failure or symptoms of coronary artery disease), as determined by medical history and physical examination. Patients with a history of cardiac disease must have a normal cardiac stress test (treadmill, echocardiogram, or myocardial perfusion scan) within the past 6 months of study entry.

3.2.11 Active infections within 48 hours of study entry

3.2.12 Systemic infection requiring chronic maintenance or suppressive therapy

3.2.13 Patients are excluded for any underlying medical or psychiatric condition, which in the opinion of the investigator, will make treatment hazardous or obscure the interpretation of adverse events, such as a condition associated with frequent rashes or diarrhea.

4.0 Patient Registration

All patients entered on any WFBCCC trial, whether treatment, companion, or cancer control, must be registered with the WFBCCC Protocol Registrar or entered into the Oncology Research Information System (ORIS) Screening Log within 24 hours of Informed Consent.

In order to ensure prompt registration of your patient:

1. Complete the Eligibility Checklist (Appendix A)
2. Complete the Protocol Registration Form (Appendix B)

3. Alert the WFUHS registrar by phone, and then send the signed Informed Consent Form, Eligibility Checklist and Protocol Registration Form to the registrar, either by fax or e-mail. Ensure that the “source documents” column of the checklist is complete and that the form is signed by either the research nurse or CRA. Patients will not be registered if forms are incomplete.

Protocol Registrar Contact Information:

PHONE (336) 713-6767
FAX (336) 713-6772
E-MAIL (registra@wakehealth.edu)

*Protocol Registration is open from 8:30 AM - 4:00 PM, Monday-Friday.

To complete the registration process, the Registrar will assign a patient study number, randomize the patient, and register the patient to the study.

5.0 Treatment Plan

5.1 Dosing

5.1.1 Leukapheresis to collect PBMC for infusion will be performed the day prior to infusion. Patients will be administered three infusions of APN401 at 50 $x10^5$ cells/kg IV, separated by 4 weeks (reference Appendix I). Leukapheresis and administration of APN401 can be delayed up to 3 weeks at the physician’s discretion. Subsequent treatment must occur 28 days after prior treatment.

5.1.2 Six patients will be treated initially. Should >1/6 subjects experience greater than grade 3 or 4 toxicity, further enrollment of study subjects and further administration of the study agent will stop pending assessment of the risk.

5.1.3 If DLT is observed in ≤ 1/6, than an additional four patients (a total of 12) will be enrolled to allow evaluation of laboratory correlates. Enrollment will be suspended if >1/6 or >3/12 patients manifest DLT

5.2 Leukapheresis

5.2.1 PBMC will be harvested from the peripheral blood by a standard 1.5 to 2.0 blood volume mononuclear cell leukapheresis.

5.2.2 Mobilization with a colony-stimulating factor is not required.

5.3 Infusion

5.3.1 Confirm patient identity: APN401 is intended solely for autologous use. Confirm the proper product has been received according to the label on the outside of the insulated polyurethane container. Prior to APN401 infusion, match the patient’s identity with the patient identifiers on the cell product disposition form and the APN401 infusion bag.

5.3.2 Confirm expiration date and time: Infusion must begin prior to the expiration date and time indicated on the cell product disposition form and Product Label. **Do not initiate infusion of expired APN401**.

5.3.3 Inspect the infusion bag: Inspect the bag for signs of leakage. Do not administer if the bag leaks. Contents of the bag will be slightly cloudy, with a cream-to-pink color. Gently mix and re-suspend the contents of the bag, inspecting for clumps and clots. Small clumps of cellular material should disperse with gentle manual mixing. Do not administer if the bag leaks during handling or if clumps remain in the bag.

5.3.4 Vital signs will be recorded at screening, prior to infusion and at 30 and 60 minutes after infusion.

5.3.5 Administer APN401 via intravenous infusion over a period of approximately 30 minutes. **Do not use a cell filter.** The entire volume of the bag should be infused.

5.3.6 Observe the patient for at least 60 minutes following each infusion.

5.4 Supportive care guidelines

5.4.1 Patients should receive *full supportive care*, including transfusions of blood and blood products, antibiotics, antiemetics, etc. when appropriate. The reason(s), the drugs and dosage, and the dates of supportive treatments should be recorded on flow sheets.

5.4.2 Patients may receive the following to abrogate APN401 infusion toxicity:
- Acetaminophen 650 mg po q 4 hr. May be given as pre-treatment at physician's discretion.
- Hydroxyzine hydrochloride or diphenhydramine may be used for generalized erythematous rash. May be given as pre-treatment at physician's discretion.
- Demerol 25-75 mg intravenously q 20 min can be administered concurrently to control chills and rigors.
- Antiemetics and antidiarrheal agents as required.

5.4.3 Treatment with *hormones or other chemotherapeutic agents* may not be administered except for steroids given for pre-existing adrenal failure; hormones administered for non-disease-related conditions (*e.g.*, insulin for diabetes). Use of dexamethasone and other steroidal antiemetic or anti-inflammatory drugs is prohibited.

5.4.4 In the event of an acute infusion reaction, the infusion may be interrupted or slowed, depending on the severity of the reaction. Appropriate medical therapy should be administered as needed. If the infusion of APN401 must be interrupted, the infusion should not be resumed if the APN401 infusion bag will be held at room temperature for more than 3 hours.

5.4.5 APN401 is released for infusion based on the microbial and sterility results from several tests: microbial contamination determination by Gram stain, endotoxin content, and mycoplasma testing. Sterility sampling will be performed by the

research technologist. APN401 is also cultured for microbial growth. The final (14-day incubation) sterility test results are not available at the time of infusion. If the sterility results become positive for microbial contamination after APN401 has been approved for infusion, the treating physician will be informed and testing to identify the microorganism as will antibiotic sensitivity testing on recovered microorganisms will be performed. The clinical investigator should evaluate the subject for any signs of infection that may be attributable the product sterility failure. Additional information from the physician in order to determine the source of contamination may be necessary. See section 8.5.1 for the reporting requirement for sterility failure.

5.5 Nursing implications

5.5.1 It is important to maintain all scheduled appointments and arrive at each appointment on time because the leukapheresis and infusions must be appropriately spaced, and the APN401 expiration time must not be exceeded.

5.5.2 If the patient is unable to receive an infusion of APN401 within the required time, the patient will need to undergo an additional leukapheresis procedure if the treatment is to be continued.

5.5.3 Counsel the patient on the importance of adhering to preparation instructions for the leukapheresis procedure, the possible side effects of leukapheresis, and post-procedure care.

5.5.4 If the patient does not have adequate peripheral venous access to accommodate the leukapheresis procedure and infusion of APN401, inform the patient about the need for a central venous catheter. Counsel the patient on the importance of catheter care. Instruct the patient to tell their doctor if they are experiencing fevers or any swelling or redness around the catheter site, because these symptoms could be signs of an infected catheter.

5.5.5 Report signs and symptoms of acute infusion reactions such as fever, chills, fatigue, breathing problems, dizziness, high blood pressure, nausea, vomiting, headache, or muscle aches.

5.5.6 Report any symptoms suggestive of a cardiac arrhythmia.

5.5.7 Inform the treating doctor if the patient is taking immunosuppressive agents.

5.6 Duration of therapy

Patients will receive protocol therapy unless:

5.6.1 Extraordinary medical circumstances: If at any time the constraints of this protocol are detrimental to the patient's health, protocol treatment should be discontinued. This includes any clinical adverse event, laboratory abnormality or intercurrent illness which, in the opinion of the investigator, indicates that continued treatment with study therapy is not in the best interest of the subject.

5.6.2 Patient withdraws consent

5.6.3 Pregnancy: All WOCBP should be instructed to contact the investigator immediately if they suspect they might be pregnant (e.g., missed or late menstrual period) at any time during study participation. Pregnancy tests should be conducted as clinically indicated.

5.6.4 Disease progression: Patients who progress clinically before week 9 will be removed from study if other therapy (surgery, radiation, chemotherapy) is needed urgently.

When a patient is removed from study, the Off-Study Form/Early Termination must be completed within seven days.

5.7 Duration of follow-up

All patients, including those who discontinue protocol therapy early, will be followed for response until progression or one year from registration, whichever occurs first.Patients will then be followed for survival for five years from the date of registration. All patients must also be followed through completion of all protocol therapy.

6.0 Study Calendar

Baseline evaluations, including imaging, are to be conducted within 4 weeks prior to initiation of treatment (Day 0). In the event that the patient's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy.

		Treatment Phase							Follow-Up Phase		Early Termination
	Pre-Study[J]	Day 0 Week 1	Day 1 Week 1	Day 15 Week 3 (± 1 day)	Day 28 Week 5 (± 3 days)	Day 29[N] Week 5	Day 56 Week 9 (± 3 days)	Day 57[N] Week 9	Day 71 Week 10 (± 3 days)	Week 18 and every 8 weeks (± 1 week)[L]	Off Study Visit/Early Termination Visit[L,M]
Leukapheresis[A]		X			X		X				
APN401[B]			X			X		X			
Informed consent	X										
Demographics	X										
Medical history	X										
Concurrent meds		X			X		X		X	X[O]	X
Vital signs[C]	X	X	X	X	X	X	X	X	X	X	X
Physical exam[Q]	X	X		X	X		X		X	X	X
Performance status	X	X		X	X		X		X	X	X
CBC w/diff[D]	X	X		X	X		X		X	X	X
Chemistries[E]	X	X		X	X		X		X	X	X
PT, PTT	X										
HIV, HBV, HCV	X										
βhCG[F]	X										
Adverse events		X	X	X	X	X	X	X	X		X
Imaging[G]	X								X[L]	X[K]	X[L]
Tumor marker[H]	X								X	X	X
Immune response[I]		X		X	X		X			X	X

A. PBMC will be harvested from the peripheral blood by a standard 1.5 to 2.0 blood volume mononuclear cell leukapheresis.
B. APN401 is infused 50 x10^5/kg IV over 30 minutes
C. Vitals signs should include blood pressure, heart rate, temperature, and respiratory rate. At the administration visits, vitals signs should also be performed before infusion and 30 and 60 minutes after the end of the infusion.
D. Hemoglobin, hematocrit, white blood cell count, and platelets. The CBC differential includes enumeration of neutrophils, lymphocytes, eosinophils, monocytes, basophils and any abnormal blood cells.
E. Serum albumin, urea or BUN, creatinine, ALT, AST, LDH, alkaline phosphatase, direct and total bilirubin, glucose, total protein, sodium, potassium, chloride, CO_2, calcium, phosphorus, and uric acid. These labs should be obtained less than 36 hours prior to the treatment.
F. Women of child bearing potential must have a negative serum or urine pregnancy test (minimum sensitivity 25 IU/L or equivalent units of HCG) at screening. A pregnancy test should also be obtained at any time if there are clinical concerns for possible pregnancy.
G. Imaging should include a chest and abdomen CT. Other types of imaging and/or imaging of other sites should be obtained as clinically indicated to document extent of cancer. If restaging imaging indicates partial or complete response, repeat confirmatory imaging should be obtained in 4 weeks (but

no sooner than 4 weeks).
H. CA19-9 in patients with pancreatic cancer, CEA in patients with colorectal cancer, and other standard-of-care tumor markers as clinically indicated. Repeat assessment not required if completed within 2 months of pre-study or early termination visit.
I. On Days 0, 28 and 56, the immune response testing will be taken from the leukapheresis product. On Day 15 and in the follow-up phase, collect 30 ml of blood (3 vials) into 10 ml EDTA tubes. If immune response labs have been collected within 4 weeks prior to the off study/ termination visit, re-collection is not required.
J. Peripheral blood laboratory studies performed in order to determine patient eligibility must be performed within 28 days of study entry. Radiographic and other tests performed in order to determine patient eligibility must be performed within 28 days of study entry.
K. Follow-up radiologic procedures used to assess outcome should be performed within 7 working days of the scheduled time.
L. Patients will be observed until objective disease progression or 1 year, whichever occurs first.
M. Off study/early termination assessments are only indicated for patients who do not complete all study visits (until week 58) /Follow-Up phase. These assessments should occur no more than 4 weeks after the discontinuation of study treatment. If the subject had not undergone imaging within the past 6 weeks re-imaging is indicated. If a patient discontinues treatment for any reason other than progression, imaging will be done every 12 weeks until progression.
N. Must occur the day following leukapheresis day.
O. Following the week 18 visit only documentation of anti-cancer treatment.
P. Adverse events will be recorded on an ongoing basis and up to 2 months following the administration of the test drug.
Q. Physical Exam may be completed +/- 2 days of visit.

7.0 Drug information

7.1 Description

The drug product, APN401, is a suspension of viable PBMCs transfected with siRNA in order to reduce Cbl-b protein levels of the respective patient (autologous administration) in saline

7.2 How supplied

Each dose of APN401 contains autologous PBMCs transfected with siRNA in order to reduce Cbl-b protein levels suspended in saline

7.3 Side effects

Infusions of APN401 have been well tolerated. Grade 3 or 4 toxicities attributable to treatment were not observed. Chills are the most common toxicity. These manifested at the completion of the 30 minute infusion and responded to meperidine. Transient lymphopenia, attributed to the leukapheresis, was also observed. Anemia, anorexia, dyspnea, fatigue, fever, hyponatremia, nausea, and myalgia have also been observed.

7.4 Storage and handling

Storage is not recommended. The infusion bag can be maintained at room temperature for up to three hours after release from the laboratory. APN401 should not be infused if maintained more than three hours at room temperature. APN401 is **not** routinely tested for transmissible infectious diseases and may transmit diseases to health care professionals handling the product. Universal precautions should be followed.

7.5 Administration

APN401 is administered at 50 $x10^5$/kg via intravenous infusion over a period of approximately 30 minutes. **Do not use a cell filter.**

7.6 Cell Processing

Cell processing is performed in the Blood and Bone Marrow Transplant Lab in the North Tower at WFBMC. The BMT lab is not accredited for this purpose and does not have responsibility for the procedures as part of this study within the laboratory. The research team, which includes the PI and an experienced technician, is responsible for the safety, purity and potency of the cellular products under protocol.

8.0 Toxicity Criteria and Adverse Event Reporting

8.1 Adverse event definition

An adverse event is any physical or clinical change or disease experienced by the subject at any time during the course of the study, whether or not considered related to the use of the study drug. This includes the onset of new illness and the exacerbation of pre-existing

conditions. Disease-related symptoms that do not increase in severity or frequency from baseline are not considered adverse events.

All toxicity should be graded according to the NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0. Adverse events will be recorded on an ongoing basis and up to 2 months following the administration of the test drug. All appropriate treatment areas should have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov).

- **'Expectedness'**: AEs can be 'Unexpected' or 'Expected' (see Section 7.3 above) for expedited reporting purposes only.

- **Attribution** of the AE:

  Definite – The AE **is clearly related** to the study treatment.
  Probable – The AE **is likely related** to the study treatment.
  Possible – The AE **may be related** to the study treatment.
  Unlikely – The AE **is doubtfully related** to the study treatment.
  Unrelated – The AE **is clearly NOT related** to the study treatment.

8.2 Dose limiting toxicity (DLT)

DLT is defined as

8.2.1 CTCAE grades 3-5 allergic reactions related to study cell infusion

8.2.2 CTCAE grades 3 and greater autoimmune reactions other than that vitiligo

8.2.3 CTCAE grades 3 and greater organ toxicity (cardiac, dermatologic, gastrointestinal, hepatic, pulmonary, renal/genitourinary or neurologic) not pre-existing or due to the underlying malignancy and occurring within 30 days of study product infusion

8.2.4 Treatment-related death within 8 weeks of the study cell infusion

8.3 Safety and toxicity reporting requirements

The Safety and Toxicity Reporting Committee (STRC) is responsible for reviewing SAEs for WFBCCC Institutional studies as outlined in Appendix C. STRC currently requires that all unexpected 4 and all grade 5 SAEs on these trials be reported to the STRC for review. Any grade 3 to 5 product infusion reactions must also be reported. All WFBCCC Clinical Research Management (CRM) staff members assisting a Principal Investigator in investigating, documenting and reporting an SAE that qualifies for STRC reporting are responsible for informing a clinical member of the STRC – as well as the entire committee – via the email notification procedure of the occurrence of an SAE.

8.4 WFUHS IRB AE reporting requirements

Any unanticipated problems involving risks to subjects or others and adverse events shall be promptly reported to the IRB, according to institutional policy. Reporting to the IRB is

required regardless of the funding source, study sponsor, or whether the event involves an investigational or marketed drug, biologic or device. Reportable events are not limited to physical injury, but include psychological, economic and social harm. Reportable events may arise as a result of drugs, biological agents, devices, procedures or other interventions, or as a result of questionnaires, surveys, observations or other interactions with research subjects.

All members of the research team are responsible for the appropriate reporting to the IRB and other applicable parties of unanticipated problems involving risk to subjects or others. The Principal Investigator, however, is ultimately responsible for ensuring the prompt reporting of unanticipated problems involving risk to subjects or others to the IRB. The Principal Investigator is also responsible for ensuring that all reported unanticipated risks to subjects and others which they receive are reviewed to determine whether the report represents a change in the risks and/or benefits to study participants, and whether any changes in the informed consent, protocol or other study-related documents are required.

Any unanticipated problems involving risks to subjects or others occurring at a site where the study has been approved by the WFUHS IRB (internal events) must be reported to the WFUHS IRB within seven calendar days of the investigator or other members of the study team becoming aware of the event.

Any unanticipated problems involving risks to subjects or others occurring at another site conducting the same study that has been approved by the WFUHS IRB (external events) must be reported to the WFUHS IRB within seven calendar days of the investigator or other members of the study team becoming aware of the event.

Any event, incident, experience, or outcome that alters the risk versus potential benefit of the research and as a result warrants a substantive change in the research protocol or informed consent process/document in order to insure the safety, rights or welfare of research subjects.

8.5 FDA reporting requirements

IND sponsors are responsible for compliance with the Federal Food, Drug, and Cosmetic Act and the implementing regulations (Title 21 of the Code of Federal Regulations). Those responsibilities include: (1) reporting any unexpected fatal or life threatening adverse experience associated with use of the drug by telephone or fax no later than 7 calendar days after initial receipt of the information [21 CFR 312.32(c)(2)]; (2) reporting any adverse experience associated with use of the drug that is both serious and unexpected in writing no later than 15 calendar days after initial receipt of the information [21 CFR 312.32(c)(1)]; and (3) submitting annual progress reports (21 CFR 312.33).

8.5.1 Sterility failure, result of investigation of the cause and any corrective actions, must be reported in an information amendment submitted to the FDA in a timely manner, within 30 calendar days after initial receipt of the positive culture test (21 CFR 312.31). If the patient experiences any serious and unexpected adverse experience that could be from administration of the sterility failure of the cellular product, then this information must reported to the FDA in an IND safety report no more than 15 calendar days after the initial receipt of the information (21 CFR 312.32).

8.5.2 If study stopping rules are met and if it is determined that the investigational drug presents an unreasonable and significant risk to subjects, all investigations are to be discontinued and the FDA, all Institutional Review Boards, and all investigators need to be notified (21 CFR 312.56(d).

9.0 Clinical Response

9.1 Definitions

Response and progression will be evaluated in this study using the new international criteria proposed by the Response Evaluation Criteria in Solid Tumors (RECIST) Committee [*JNCI* 92(3):205-216, 2000]. Changes in only the largest diameter (unidimensional measurement) of the tumor lesions are used in the RECIST criteria. Note: lesions are either measurable or non-measurable using the criteria provided below. The term “evaluable” in reference to measurability will not be used because it does not provide additional meaning or accuracy.

9.1.1 Measurable disease: Measurable lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as ≥20 mm with conventional techniques (PET, CT, MRI, x-ray) or as ≥10 mm with spiral CT scan. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

9.1.2 Non-measurable disease: All other lesions (or sites of disease), including small lesions (longest diameter <20 mm with conventional techniques or <10 mm using spiral CT scan), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonis, inflammatory breast disease, abdominal masses (not followed by CT or MRI), and cystic lesions are all non-measurable.

9.1.3 Target lesions: All measurable lesions up to a maximum of five lesions per organ and 10 lesions in total representative of all involved organs should be identified as target lesions and recorded and measured at baseline. Target lesions should be selected on the basis of their size (lesions with the longest diameter) and their suitability for accurate repeated measurements (either by imaging techniques or clinically). A sum of the longest diameter (LD) for all target lesions will be calculated and reported as the baseline sum LD. The baseline sum LD will be used as reference by which to characterize the objective tumor response.

9.1.4 Non-target lesions: All other lesions (or sites of disease) should be identified as non-target lesions and should also be recorded at baseline. Non-target lesions include measurable lesions that exceed the maximum numbers per organ or total of all involved organs as well as non-measurable lesions. Measurements of these lesions are not required but the presence or absence of each should be noted throughout follow-up.

9.2 Measurable disease guidelines

All measurements should be taken and recorded in metric notation using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than four weeks before the beginning of the treatment.

- The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation is preferred to evaluation by clinical examination when both methods have been used to assess the antitumor effect of a treatment.

- Clinical lesions. Clinical lesions will only be considered measurable when they are superficial (e.g., skin nodules and palpable lymph nodes). In the case of skin lesions, documentation by color photography, including a ruler to estimate the size of the lesion, is recommended.

- Chest X-ray. Lesions on chest x-ray are acceptable as measurable lesions when they are clearly defined and surrounded by aerated lung. However, CT is preferable.

- Conventional CT and MRI. These techniques should be performed with cuts of 10 mm or less in slice thickness contiguously. Spiral CT should be performed using a 5 mm contiguous reconstruction algorithm. This applies to tumors of the chest, abdomen, and pelvis. Head and neck tumors and those of extremities usually require specific protocols.

- PET-CT. At present, the low dose or attenuation correction CT portion of a combined PET-CT is not always of optimal diagnostic CT quality for use with RECIST measurements. However, if the site can document that the CT performed as part of a PET-CT is of identical diagnostic quality to a diagnostic CT (with IV and oral contrast), then the CT portion of the PET-CT can be used for RECIST measurements and can be used interchangeable with conventional CT in accurately measuring cancer lesion over time. Note, however, that the PET portion of the CT introduces additional data which may bias an investigator if it is not routinely or serially performed.

- Ultrasound (US). When the primary endpoint of the study is objective response evaluation, US should not be used to measure tumor lesions. It is, however, a possible alternative to clinical measurements of superficial palpable lymph nodes, subcutaneous lesions, and thyroid nodules. US might also be useful to confirm the complete disappearance of superficial lesions usually assessed by clinical examination.

- Endoscopy, laparoscopy. The utilization of these techniques for objective tumor evaluation has not yet been fully and widely validated. Their uses in this specific context require sophisticated equipment and a high level of expertise that may only be available in some centers. Therefore, the utilization of such techniques for objective tumor response should be restricted to validation purposes in reference centers. However, such techniques can be useful to confirm complete pathological response when biopsies are obtained.

- Tumor markers. Tumor markers alone cannot be used to assess response. If markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response.

- Cytology, histology. These techniques can be used to differentiate between partial responses (PR) and complete responses (CR) in rare cases (e.g., residual lesions in tumor types such as germ cell tumors, where known residual benign tumors can remain).

9.3 Response criteria

9.3.1 Evaluation of target lesions

- Complete Response (CR): Disappearance of all target lesions

- Partial Response (PR): At least a 30% decrease in the sum of the longest diameter (LD) of target lesions, taking as reference the baseline sum LD

- Progressive Disease (PD): At least a 20% increase in the sum of the LD of target lesions, taking as reference the smallest sum LD recorded since the treatment started or the appearance of one or more new lesions

- Stable Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum LD since the treatment started

9.3.2 Evaluation of non-target lesions

- Complete Response (CR): Disappearance of all non-target lesions and normalization of tumor marker level

- Incomplete Response/Stable Disease (SD): Persistence of one or more non-target lesion(s) or/and maintenance of tumor marker level above the normal limits
- Progressive Disease (PD): Appearance of one or more new lesions and/or unequivocal progression of existing non-target lesions

- Although a clear progression of “non-target” lesions only is exceptional, in such circumstances, the opinion of the treating physician should prevail, and the progression status should be confirmed at a later time by the review panel (or study chair).

9.3.3 Evaluation of best overall response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria (see section 9.4.1).

Target Lesions	Non-target Lesions	New Lesions	Overall Response
CR	CR	No	CR
CR	Incomplete response/SD	No	PR
PR	Non-PD	No	PR
SD	Non-PD	No	SD
PD	Any	Yes or No	PD
Any	PD	Yes or No	PD
Any	Any	Yes	PD

Note: Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration." Every effort should be made to document the objective progression, even after discontinuation of treatment. In some circumstances, it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) before confirming the complete response status.

9.4 Confirmatory measurement/duration of response

9.4.1 Confirmation: To be assigned a status of PR or CR, changes in tumor measurements must be confirmed by repeat assessments that should be performed four weeks after the criteria for response are first met. In the case of SD, follow-up measurements must have met the SD criteria at least once after study entry at a minimum interval of eight weeks.

9.4.2 Duration of overall response: The duration of overall response is measured from the time measurement criteria are met for CR or PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that recurrent disease is objectively documented.

9.4.3 Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started.

10.0 Laboratory Correlates

10.1 Sample collection

Peripheral blood will be obtained at baseline prior to the first infusion, on day 29 and 57 (four and eight weeks after the infusion) and every eight weeks thereafter. Twenty (20) ml of blood is collected in two 10ml EDTA-containing vacutainers and sent to Dr. Triozzi's laboratory, Room 5029, Hanes Building.

10.2 **Immune Response Outcomes**

All research laboratory procedures will be performed per manufacturer's recommendations with amendments (if needed) as necessary to adapt the protocol to the current applications. Procedures and results will be recorded in a laboratory notebooks but also recorded on Appendices L through N.

10.2.1 PMBC IFNγ production in vitro (R&D Systems ELISA) in response to anti-CD3 plus anti-CD28 antibody (BD) (Appendix L)

10.2.2 PMBC IFNγ production in vitro (R&D Systems ELISA) in response NY-ESO, survivin, and telomerase peptide mixtures (Proimmune Promix) (Appendix L)

10.2.3 Frequency (flow cytometry) of CD4+, CD8+, CD4+FoxP3+ regulatory, ICOS+CD4+ and ICOS+CD8+ effector, and CD45RO+CD4+ memory T cells and CD3-CD56+ NK cells and CD19 B cells (Appendix M)

10.2.4 Neutrophil to lymphocyte ratio (Appendix N)

11.0 Statistical Considerations

11.1 **Study Outcomes**

11.1.1 **Primary Outcome**

- Rate of Toxicities when patients are administered APN401 for one year

11.1.2 **Secondary Outcome**

- To determine the immunologic effects of multiple infusions of APN401 through monitoring immune response in vitro by measuring IFN production, frequency of immune cells, and neutrophil to lymphocyte ratio for one year.

- To document clinical response and survival using from RECIST criteria and progression free survival and overall survival for five years, respectively.

11.1 Study parameters

The primary goals are to determine the toxicities and to determine effects on immune response. The secondary goal is to document clinical effects.

11.2 Sample size

A total of up to 12 patients will be treated to allow evaluation of laboratory correlates.

11.3 Analyses

11.3.1 Study parameters

The primary goal of this study is to estimate the rate of toxicities. The secondary goals include determining the effects of treatment on immune response and also to examine clinical effects (response (from RECIST criteria), progression free survival and overall survival)

11.3.2 Sample size

A total of up to 12 patients will be treated to allow evaluation of laboratory correlates. The design allows for an interim analysis to occur after the first 6 patients are enrolled. If 1 or less experience a grade 3-4 toxicity then 4 more will be enrolled, if 3 or less experience a grade 3-4 toxicity the agent will not be considered too toxic. With this sample size, during the first stage of the trial, if the true toxicity rate is 10% (or less) the trial will stop with 11% probability, whereas if the true toxicity rate is 40% (or higher) then the trial will stop with 76.7% probability. Likewise, if the trial continues to 12 patients, if the true toxicity rate is 10% or less the probability of observing more than 3 toxicities is 1.2% and if the true toxicity rate is 40% or more then the probability of observing more than 3 toxicities is 62%.

11.4 Analyses

Adverse events will be categorized by organ system and severity and summarized as frequency counts and percentages. Other categorical data, such as clinical response, will be summarized similarly. Measured data, such as immune response, will be summarized as medians and ranges. All tests of statistical significance will be 2-sided and p-values will generally not be adjusted for multiple comparisons.

11.4.1 Toxicity: APN401 will be considered tolerable if ≤1/6 or if overall ≤3/12 patients (i.e. an observed toxicity rate of at most 30%) experience grade 3-4 toxicity. With this design the likelihood of stopping early and rejecting the therapy as too toxic is ≤11.4% if the true grade 3-4 toxicity rate is ≤10% and >76.7% if it is >40%. The overall likelihood of declaring APN401 too toxic is ≤1.2% and >62%, respectively in these scenarios.

11.4.2 Immune response: APN401 will be considered immunologically active if PBMC IFN-γ production in response to anti-CD3/28 stimulation or to one or more tumor antigens is significantly increased or if neutrophil to lymphocyte ratio is significantly decreased post-therapy (week 9) compared to pre therapy. Immune responses will be summarized as medians and ranges. The effects of treatment on these markers individually will be analyzed using paired t-tests or the non-parametric counterpart. With 12 patients there will be statistical power equal to 80% to detect 1 1 standard deviation change in these markers following treatment. All tests will be 2-sided and $P < 0.05$ will be considered significant.

11.4.3 Clinical response and survival: Clinical response will be assessed by RECIST and summarized as frequency counts and percentages. Progression-free survival and overall survival are calculated from the initial infusion to confirmation of progression or death. Exploratory survival plots will be estimated using the Kaplan Meier approach and median PFS and OS will be estimated if enough events occur.

11.5 Interim Analysis

As described above an interim analysis will occur after 6 patients are enrolled

11.6 Accrual Rate

Accrual is anticipated to be 1-2 patients per month. However, since accrual will stop after the first 6 patients are enrolled, the total time for the study is anticipated 12-15 months.

12.0 Data Management Procedures

Informed Consent Form	ORIS
Subject Eligibility Checklist (Appendix A)	ORIS
Protocol Registration Form (Appendix B)	ORIS
Concurrent Medications Form	Sponsor Database
Specimen Accountability Log (Appendix D)	Laboratory Use
RECIST Measurement Form (Appendix E)	ORIS
Response Criteria (Appendix F)	ORIS
Survival Data	Sponsor Database
Off-Study Form	Sponsor Database
Interferon Production Data Form (Appendix H)	Laboratory use
Frequency Count Data Form (Appendix I)	Laboratory use
Neutrophil/Lymphocyte Ratio Form (Appendix J)	Laboratory use

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Appendix A – Subject Eligibility Checklist

IRB Protocol No. 00041173	**CCCWFU Protocol No. 03716**
Study Title: Safety and Immunologic Activity of Multiple Infusions of APN401	
Principal Investigator: Pierre L. Triozzi, MD	

Inclusion Criteria (as outlined in study protocol)	**Criteria is met**	**Criteria is NOT met**	**Source Used to Confirm ***
3.1.1 Patients with histologically confirmed inoperable, recurrent or metastatic malignant solid tumors, deemed incurable, and who have either: • failed to respond to standard therapy or • for whom no standard therapy is available or • refuse to receive standard therapies The study is intended to enroll patients with pancreatic and colorectal cancer. Patients with other types of solid tumors will require approval by the Principal Investigator.	☐	☐	
3.1.2 Measurable disease as defined by RECIST (see Section 9.0)	☐	☐	
3.1.3 Patients with treated, stable, and asymptomatic brain metastases are eligible	☐	☐	
3.1.4 Patients on every 2, 3 or 4 week systemic therapy programs must be off the treatment program at least 2, 3, or 4 weeks, respectively, and must have recovered from any clinically significant toxicity experienced. Patients on weekly or daily systemic therapy programs and patients receiving radiation must be at least 1 week since treatment and recovered from any clinically significant toxicity experienced. Must be at least 4 weeks and have recovered from major surgery	☐	☐	
3.1.5 Age ≥ 18 years	☐	☐	
3.1.6 ECOG performance status 0 or 1	☐	☐	

3.1.7 Patients must have the following lab values obtained < 4 weeks prior to starting treatment: • WBC ≥ 3000/μL • Platelets ≥ 100,000/μL • Hematocrit ≥ 28% • Creatinine ≤ 1.6 mg/dL • AST and ALT < 2.5 x upper limit of normal • Bilirubin ≤ 1.6 mg/dL (except patients with Gilbert's Syndrome, who must have a total bilirubin less than 3.0 mg/dL) • Albumin ≥ 3.0 g/dL • INR ≤ 1.5	☐	☐	
Exclusion Criteria (as outlined in study protocol)	**Criteria NOT present**	**Criteria is present**	**Source Used to Confirm ***
3.2.1 Women must not be pregnant or breastfeeding due to unknown effects of the study treatments on the unborn fetus. All women of childbearing potential must have a blood test within 72 hours prior to randomization to rule out pregnancy. Women of childbearing potential and sexually active males must be strongly advised to use an accepted and effective method of contraception. Women of childbearing potential (WOCBP) must be using an adequate method of contraception to avoid pregnancy throughout the study and for 26 weeks after the last dose of investigational product, in such a manner that the risk of pregnancy is minimized. Sexually mature females who have not undergone a hysterectomy or who have not been postmenopausal naturally for at least 24 consecutive months (i.e., who have had menses at some time in the preceding 24 consecutive months) are considered to be of childbearing potential. Women who are using oral contraceptives, other hormonal contraceptives (vaginal products, skin patches, or implanted or injectable products), or mechanical products such as an intrauterine device or barrier methods (diaphragm, condoms, spermicides) to prevent pregnancy, or are practicing abstinence or where their partner is sterile (e.g., vasectomy) should be considered to be of childbearing potential.	☐	☐	
3.2.2 Untreated, progressing, or symptomatic brain metastases	☐	☐	

3.2.3 Autoimmune disease, as follows: Patients with a history of inflammatory bowel disease are excluded as are patients with a history of symptomatic disease (e.g., rheumatoid arthritis, systemic progressive sclerosis [scleroderma], systemic lupus erythematosus, autoimmune vasculitis [e.g., Wegener's granulomatosis]). Patients with motor neuropathy considered of autoimmune origin (e.g., Guillain-Barre Syndrome and myasthenia gravis) are excluded. Patients with a history of autoimmune thyroiditis are eligible if their current thyroid disorder is treated and stable with replacement or other medical therapy.	☐	☐	
3.2.4 Any other malignancy from which the patient has been disease-free for less than 2 years, with the exception of adequately treated and cured basal or squamous cell skin cancer, superficial bladder cancer or carcinoma *in situ* of the cervix.	☐	☐	
3.2.5 Other ongoing systemic therapy for cancer, including any other experimental treatment. These include concomitant therapy with any of the following: IL-2, interferon, ipilimumab, pembrolizumab, nivolumab or other immunotherapy; cytotoxic chemotherapy; and targeted therapies.	☐	☐	
3.2.6 Ongoing requirement for an immunosuppressive treatment, including the use of glucocorticoids or cyclosporine, or with a history of chronic use of any such medication within the last 4 weeks before enrolment. Patients are excluded if they have any concurrent medical condition that requires the use of systemic steroids (the use of inhaled or topical steroids is permitted).	☐	☐	
3.2.7 Infection with HIV. Due to the mechanism of action of APN401, activity and side effects in an immune compromised patient are unknown.	☐	☐	
3.2.8 Active infection with Hepatitis B. Active or chronic infection with Hepatitis C.	☐	☐	
3.2.9 Clinically significant pulmonary dysfunction, as determined by medical history and physical examination. Patients with a history of pulmonary dysfunction must have pulmonary function tests with a $FEV_1 \geq 60\%$ of predicted and a DLCO ≥ 55% (corrected for hemoglobin)	☐	☐	

3.2.10 Clinically significant cardiovascular abnormalities (e.g., congestive heart failure or symptoms of coronary artery disease), as determined by medical history and physical examination. Patients with a history of cardiac disease must have a normal cardiac stress test (treadmill, echocardiogram, or myocardial perfusion scan) within the past 6 months of study entry.	☐	☐	
3.2.11 Active infections within 48 hours of study entry	☐	☐	
3.2.12 Systemic infection requiring chronic maintenance or suppressive therapy	☐	☐	
3.2.13 Patients are excluded for any underlying medical or psychiatric condition, which in the opinion of the investigator, will make treatment hazardous or obscure the interpretation of adverse events, such as a condition associated with frequent rashes or diarrhea.	☐	☐	

This subject is ☐ eligible / ☐ ineligible for participation in this study.

ORIS Assigned PID: _____

Research professional confirming eligibility:

Signature_____ Date: _____

Treating Physician:

Signature_____ Date: _____

Principal Investigator**:

Signature_____ Date: _____

* Examples of source documents include clinic note, pathology report, laboratory results, etc. When listing the source, please specifically state which document in the medical record was used to assess eligibility. Please also include the date on the document. Example: “Pathology report, 01/01/14” or “Clinic note, 01/01/14”

**Principal Investigator signature can be obtained following registration if needed

Appendix B – Protocol Registration Form

DEMOGRAPHICS

Patient: Last Name: _____ First Name: _____

MRN: ___ ___ ___ ___ ___ ___ ___ ___ DOB (mm/dd/yy): ___ ___ / ___ ___ / ___ ___

Zip Code: ___ ___ ___ ___ ___

SEX: ☐ Male ☐ Female Ethnicity (choose one): ☐ Hispanic ☐Non-Hispanic

Race (choose all that apply): ☐ WHITE ☐BLACK ☐ ASIAN ☐ PACIFIC ISLANDER ☐ NATIVE AMERICAN

Height: ___ ___.___ inches Weight: ___ ___ ___.___lbs.(actual)

Surface Area: ___ ___.___m^2

Primary Diagnosis:

_____

Date of Diagnosis: ___ ___ /___ ___/___ ___

PROTOCOL INFORMATION

Date of Registration: ___ ___ / ___ ___ / ___ ___

MD Name (last) : _____

Date protocol treatment started: ___ ___ / ___ ___ / ___ ___

Informed written consent: (consent must be signed prior to registration) ☐ YES ☐ NO

Date Consent Signed: ___ ___ / ___ ___ / ___ ___

PID # (to be assigned by ORIS): ___ ___ ___ ___ ___ ___ ___ ___

Protocol Registrar can be contact by calling 336-713-6767 between 8:30 AM and 4:00 PM, Monday – Friday.

Completed Eligibility Checklist and Protocol Registration Form must be hand delivered, faxed or e-mailed to the registrar at 336-7136772 or registra@wfubmc.edu.

Appendix C – STRC Reporting Guidelines

Safety and Toxicity Review Committee (STRC; previously known as CROC) Serious Adverse Event (SAE) Notification SOP	**Date: 11/17/2016**

Mandatory STRC SAE Reporting Requirements

This document describes STRC reporting and use of the electronic submission form that is submitted **for unexpected grade 4 and any grade 5 (death during protocol intervention) SAEs on CCCWFU Institutional interventional trial patients**. There are multiple entities that require reporting of SAEs. Each entity has different rules for what is reported, and how it is reported.

Rules used by other entities (Institutional Review Board (IRB), AdEERS, MedWatch, etc.) should NOT be used to evaluate whether an event should be reported to STRC. Only the rules for reporting described in this document should be considered.

As defined in the NCI Data Table 4 reporting guidelines, **CCCWFU Institutional Interventional studies covered by these reporting requirements are defined as: In-house, internally reviewed trials, including those collaborative studies conducted with industry sponsorship in which the center is a primary contributor to the design, implementation, and monitoring of the trial, or participation in a multi-site trial initiated by an institutional investigator at another center.** Institutional trials are almost always authored by a researcher here at CCCWFU. Institutional protocols are labeled NCI Code="I" for Institutional on the protocol screen in ORIS. Cooperative group protocols are **not** considered Institutional, but Research Base trials **are** classified as Institutional.

The STRC is responsible for reviewing SAEs for CCCWFU Institutional Interventional studies, as defined above. STRC currently requires that unexpected grade 4 and all grade 5 SAEs on these trials be reported to the STRC for review. All Clinical Protocol and Data Management (CPDM) staff members assisting a PI in documenting and reporting an SAE that qualifies for STRC reporting are responsible for informing a clinical member of the STRC by phone (or in-person), followed by informing the entire committee via the required email notification.

THESE REPORTING REQUIREMENTS APPLY TO any faculty or staff member on the study team for a CCCWFU Institutional Interventional trial. Once an event is observed, it is the responsibility of the person who observed the event to be sure that it is reported.

What is considered an SAE under this mandatory procedure?
Any **unexpected grade 4** event and **all grade 5 events** (death during protocol intervention) should be reported. These events should be reported if they occur while a patient is on study treatment or if they occur within 30 days of last study treatment (even if patient begins a new treatment during the 30 days). This window of 30 days should be the standard window to be used in all protocols unless a specific scientific rationale is presented to suggest that a shorter window can be used to identify events. In addition, if it is not clear whether the Grade 4 is unexpected it should be reported.

Top of Form
Bottom of Form

Table 1: Summary of STRC Reporting Requirements for Institutional Pilot, Phase 1, Phase 2 and Phase 3 Interventional Trials

STRC reporting may not be appropriate for specific expected adverse events for protocols. In those situations the adverse events that will not require STRC reporting **must be specified in the text of the approved protocol.**

STRC notification responsibilities of the person handling the reporting/documenting of the SAE:
Make a phone call (or speak in person) to the appropriate clinical member of the STRC as listed below (page if necessary)—see note 2 below

	ADVERSE EVENT					
	Grade 1, Grade 2, Grade 3		**Grade 4**		**Grade 5**	
	Unexpected	**Expected**	**Unexpected**	**Expected**	**Unexpected**	**Expected**
Unrelated	Not Required	Not Required	**REPORT TO STRC**	Not Required	**REPORT TO STRC**	**REPORT TO STRC**
Unlikely	Not Required	Not Required	**REPORT TO STRC**	Not Required	**REPORT TO STRC**	**REPORT TO STRC**
Possible	Not Required	Not Required	**REPORT TO STRC**	Not Required	**REPORT TO STRC**	**REPORT TO STRC**
Probable	Not Required	Not Required	**REPORT TO STRC**	Not Required	**REPORT TO STRC**	**REPORT TO STRC**
Definite	Not Required	Not Required	**REPORT TO STRC**	Not Required	**REPORT TO STRC**	**REPORT TO STRC**

Submit the STRC Notification Form WITHIN 24 HOURS of first knowledge of the event. This form is found at either the ORIS main menu page or by going to http://ccc.wfubmc.edu/oris/strc.aspx.
This will ensure that all persons that need to be made aware of the event (i.e., study team members and STRC members) will be notified; remember to file a copy of your confirmation. (Form instructions will walk you through the required fields, consult the help page for further instructions.)
Ensure that you document that the appropriate person(s) on the STRC has been contacted. This documentation is placed on the STRC Notification form described above.
Follow up with/update the clinical member(s) of STRC regarding any new developments or information obtained during the course of the SAE investigation and reporting process.

Elements to complete the electronic STRC form:
Please use 'reply to All' when responding with one of these terms: Definite, Probable, Possible, Unlikely, or Unrelated
Patient ID (ORIS PID)
Patient Name
Patient MR#
CCCWFU(ORIS) Study Number
Title
PI Name
PI Contact Number
PI Comments
STRC Clinician notified by Phone
Notified Date
Notified Time
STRC Clinician Comments
Category [This is the Grade – Either Unexpected Grade 4 or Grade 5 should be entered]
Additional Information (IRB Reporting)(after discussion with PI or STRC Clinician
Is This Event Related to Protocol Treatment?
Is Suspension of the Protocol Needed?

Any Changes to Consent or Protocol Needed?
Was Nature or Severity of Event Unexpected?
Date of the event.
Brief description (include brief clinical history relevant to this event, including therapies believed related to event).
Date of Last Dose before event
Relevant tests/labs.
Other Relevant Treatment Information
Other Comments/Notes (include regimen of chemo and dates the patient received them if known).
Cc (email) (include treating Physician; separate email list with comma",")
Your Name
Your Email
Confirm Your Email

The Clinical Members of STRC to Notify by Phone or Page:
Bayard Powell, MD – Director-at-Large, CCCWFU; Section Head, Hematology/Oncology
6-7970 / 6-2701 / Pager 336-806-9308
Glenn Lesser, MD – Hematology Oncology 6-9527 / 6-7972 / Pager 336-806-8397
Stefan Grant, MD, JD-Hematology Oncology 3-5172/ 6-5772 Pager 336-806-6453
Jimmy Ruiz, MD-Hematology Oncology 6-0230/ Pager 336-806-9710
Kathryn Greven, MD – Vice Chair – Radiation Oncology
3-3600 / 3-6505 / Pager 336-806-8314
Marissa Howard-McNatt, MD – General Surgery 6-0545 / Pager 336-806-6438
Mercedes Porosnicu, MD- Hematology Oncology 6-7980 / 6-0230 / Pager 336-806-9150
Definition of Unavailable:
As a general guideline if the first clinician that is contacted does not respond to the phone call or page within a reasonable amount of time, then initiate contact with their backup. Give the back-up a reasonable amount of time to respond to a phone call or page before contacting another member. This is a general guideline. You must use your best judgment as a clinical research professional given the time of day, severity of the SAE, and other circumstances as to when it is appropriate to contact backup clinicians. If the event occurs near the end of day, then leave messages (voice or email) as appropriate and proceed with submitting your STRC notification form. The important criteria is that you have taken reasonable steps to notify and document that you have initiated some type of contact to one or more of the clinical members of STRC.

STRC CLINICAN RESPONSIBILITY:
It is the responsibility of the STRC clinician to review all reported events, evaluate the events as they are reported; and communicate a response to the Investigator, event reporter and the members of STRC. The review will include but not be limited to the information reported; there may be times when additional information is needed in order for an assessment to be made and further communication directly with the investigator may be warranted. STRC reserves the right to agree with the investigator's assessment if STRC does not agree with the investigator. STRC reserves the right to suspend the trial pending further investigation.
Is there any immediate danger or harm that could be present for a future patient based on the information provided in the STRC report – and if so an immediate suspension of enrollment should take place.

AMENDMENTS TO PREVIOUS REPORTS
If you are not able to supply all pertinent information with the initial submission, once the additional information is available **do not submit a new report**. Go to the original email that was received by STRC and others "reply to all" and entitle your email "**Amendment** for (list date of event and patient ID) this will avoid duplications of the same event. List the additional information which you are reporting.

Acronyms and Definitions

STRC-Safety and Toxicity Review Committee
SAE-Serious Adverse Event
IRB-Institutional Review Board
CCCWFU-Comprehensive Cancer Center Wake Forest University
ORIS-Oncology Research Information System
NCI-National Cancer Institute
CPDM-Clinical Protocol and Data Management
Interventional Trials-Therapeutic Level 1 and Level 2 trials
Therapeutic Level 1-A cancer treatment protocol aimed at directly treating/curing the patient's cancer.
Therapeutic Level 2-A therapeutic protocol not cancer treatment involves clinical activity to treat symptoms, improve the patient's quality of life, or prevent cancer.

Appendix D – Specimen Accountability Log

Protocol WFBCCC 03716: Safety and Immunologic Activity of Multiple Infusions of APN401							
Subject Identifier	**Description of Specimen**	**Date and Time of Collection**	**Visit Number**	**Storage Location**	**Date and Time of Shipping/ Release**	**Name of Person Specimen Released to**	**Initials**

Appendix E – RECIST Measurement Form

TUMOR RESPONSE WORKSHEET (Modified RECIST) **CCCWFU # _____**

Patient Name: _____ **Patient MRN: _____**

Has patient had localized RT @ baseline? □ No □ Yes If yes, which lesions? :_____

Target Lesions

	Lesion	Site	Imaging *(ie,CT, MRI)*	Baseline Date: *(Se, Im)*	Cycle_____ Date: *(Se, Im)*	Cycle_____ Date: *(Se, Im)*	Cycle_____ Date: *(Se, Im)*	Cycle_____ Date: *(Se, Im)*
TARGET Lesions	01			mm	mm	mm	mm	mm
	02			mm	mm	mm	mm	mm
	03			mm	mm	mm	mm	mm
	04			mm	mm	mm	mm	mm
	05			mm	mm	mm	mm	mm
	Sum of Diameters			mm	mm	mm	mm	mm
	% Change (% Δ) from Baseline or Nadir* & absolute value (AbV)			NA / NA	% Δ / AbV			
	Target Lesion Response			**N/A**				

Non-Target Lesions

	Lesion	Site	Imaging	Baseline	Cycle_____	Cycle_____	Cycle_____	Cycle_____
NON-TARGET Lesions	01							
	02							
	03							
	04							
	05							
	Non-Target Lesion Response			**N/A**				

	New Lesions							
New	1			N/A				
	2			N/A				
	3			N/A				

Overall Tumor Response		Cycle_____	Cycle_____	Cycle_____	Cycle_____
Radiologist Signature/Date:					
Treating Physician Signature/Date:					
PI Signature:					

***Terms & Calculations**

Baseline: *The set of data collected prior to randomization*

Nadir: *The lowest point*

$$\frac{\text{Current SLD} - \text{Baseline or Nadir SLD}}{\text{Baseline or Nadir SLD}} \times 100\%$$

Appendix F – Response Criteria

ORIS Assigned PID: _____ Date Completed: ____ ___ / ___ ___ / ___ ___

Study Visit:
- ☐ Week 10, Day 71 (± 3 Days)
- ☐ Week 18 (± 1 week)
- ☐ Week 26 (± 1 week)
- ☐ Week 34 (± 1 week)
- ☐ Week 42 (± 1 week)
- ☐ Week 50 (± 1 week)
- ☐ Week 58 (± 1 week)
- ☐ Off Study
- ☐ Other visit: (please specify) _____

Evaluation of target lesions

☐ Complete Response (CR): Disappearance of all target lesions

☐ Partial Response (PR): At least a 30% decrease in the sum of the longest diameter (LD) of target lesions, taking as reference the baseline sum LD

☐ Progressive Disease (PD): At least a 20% increase in the sum of the LD of target lesions, taking as reference the smallest sum LD recorded since the treatment started or the appearance of one or more new lesions

☐ Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum LD since the treatment started

Evaluation of non-target lesions

☐ Complete Response (CR): Disappearance of all non-target lesions and normalization of tumor marker level

☐ Incomplete Response/Stable Disease (SD): Persistence of one or more non-target lesion(s) or/and maintenance of tumor marker level above the normal limits

☐ Progressive Disease (PD): Appearance of one or more new lesions and/or unequivocal progression of existing non-target lesions

Although a clear progression of “non-target” lesions only is exceptional, in such circumstances, the opinion of the treating physician should prevail, and the progression status should be confirmed at a later time by the review panel (or study chair).

Treating Physician Signature: _____ Date: ___ ___ / ___ ___ / ___ __

PI Signature: _____ Date: ___ ___ / ___ ___ / ___ ___

BEST OVERALL Response Criteria for

ORIS Assigned PID: _____ Date Completed: ____ ___ / ___ ___ / ___ ___

Study Visit:

- ☐ Week 10, Day 71 (± 3 Days)
- ☐ Week 18 (± 1 week)
- ☐ Week 26 (± 1 week)
- ☐ Week 34 (± 1 week)
- ☐ Week 42 (± 1 week)
- ☐ Week 50 (± 1 week)
- ☐ Week 58 (± 1 week)
- ☐ Off Study
- ☐ Other visit: (please specify) _____

Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria (see section 9.4.1).

Target Lesions	Non-target Lesions	New Lesions	Overall Response
CR	CR	No	CR
CR	Incomplete response/SD	No	PR
PR	Non-PD	No	PR
SD	Non-PD	No	SD
PD	Any	Yes or No	PD
Any	PD	Yes or No	PD
Any	Any	Yes	PD

☐ Complete Response (CR): Disappearance of all target lesions

☐ Partial Response (PR): At least a 30% decrease in the sum of the longest diameter (LD) of target lesions, taking as reference the baseline sum LD

☐ Progressive Disease (PD): At least a 20% increase in the sum of the LD of target lesions, taking as reference the smallest sum LD recorded since the treatment started or the appearance of one or more new lesions

☐ Disease (SD): Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum LD since the treatment started

Note: Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having "symptomatic deterioration." Every effort should be made to document the objective progression, even after discontinuation of treatment. In some circumstances, it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) before confirming the complete response status.

Confirmatory Measurement/Duration of Response

Confirmation: To be assigned a status of PR or CR, changes in tumor measurements must be confirmed by repeat assessments that should be performed four weeks after the criteria for response are first met. In the case of SD, follow-up measurements must have met the SD criteria at least once after study entry at a minimum interval of eight weeks.

Duration of overall response: The duration of overall response is measured from the time measurement criteria are met for CR or PR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that recurrent disease is objectively documented.

Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started.

Treating Physician Signature: _____ Date: ___ ___ / ___ ___ / ___ __

PI Signature: _____ Date: ___ ___ / ___ ___ / ___ ___

Appendix G – Research Study Product Chain of Custody- Apheresis/ BMT Lab

Collection of Study Product in Apheresis:

I. Identification of Research Study Participant in Apheresis

a. Identification

- Patient(s) is/are scheduled for PBMC collection by Research RN or coordinators as ordered by PI (Dr. Triozzi)
- Coordinators notify Apheresis staff of date of collection.
- Apheresis staff validates that the patient is on the proper associated research protocol (CCCWFU03716) in the EMR
- Once study status is verified the apheresis staff enter the apheresis collection plan into EMR
- Dr. Triozzi reviews and signs EMR Apheresis orders
- Patients' name is placed on Apheresis calendar and designated bed/cubicle assigned and protocol apheresis orders reviewed.
- Two registered nurses are assigned to each patient the day prior to collection
    - 1) Collection (CRN)-responsible for assigned patient and collection procedure including: labeling of product, blood tubes, Apheresis Unit\ Cellular Product Hand-off Log, and care of patient during collection of product,
    - 2) Verifier (VRN) - responsible for confirming and validating accuracy of labeling of ISBT 128 Labeling of collection bag and Related Samples (Apheresis) Product, blood tubes, and Apheresis Unit Cellular Product Hand-off Log as performed by the collection RN.
- Time Out is performed at the bedside within the confines of the cubicle space in full view and earshot of the patient.
- The CRN and VRN: Verbally and visually identify patient and verify that the patient's Name, MRN, and DOB match the protocol collection orders.

b. Labeling

- Research Protocol Specific Label provided by BMT Processing Lab: ISBT 128 research specific label placed on product bag following completion of patient and protocol order verification; both CRN and VRN sign and date the ISBT 128 product label(s) with a validated permanent marker.
- CRN then exposes adhesive backing of label and securely applies ISBT 128 product Label to the Spectra Optia IDL set collection bag. Product collection bag(s) is labeled in full view of patient and VRN, ensuring correct patient collection bag has been labeled

c. Collecting

• Collection is performed per research protocol orders. Collection via Spectra Optia Apheresis System

- Collection performed with Optia IDL set using continuous mononuclear cell collection with whole blood processed with goal of 1.5 – 2.0 TBV or greater to obtain at least 200 mL of product, collection rate of 1.0 ml/min.

d. Chain of Custody
   - Research RN/coordinator is contacted prior to completion of collection and given an exact time procedure will be complete.
   - Research RN/Coordinator coordinates the product retrieval time with Dr. Triozzi or Research designee to ensure that they are on the apheresis unit to retrieve the product from the apheresis RN immediately upon disconnection from Optia machine.
   - Study Principal Investigator (PI) is on unit to receive collected product and completes the cellular product hand off log signaling the end of the Apheresis Unit chain of custody and the assumption of the Research Protocol for subsequent chain of custody procedure adherence.
   - The research personnel then remove the product from the apheresis unit as deemed appropriate by Dr. Triozzi.

II. Minimum Processing Standards within a shared space

BMT lab Requirements:

- A meeting of all individuals including the requestors will be arranged to discuss each section of the BMT program's involvement in the research project.
- For BMT lab, discussion will include use of reagents, solutions, equipment, space, storage, time, days of the week needed for product processing, training needed for the research techs.
- Donors must be negative for infectious disease markers (IDMs)
- Transfections with viral, bacterial or any infectious agents will not be allowed in the BMT lab- Changed to Transfection will be performed in an assigned biologic safety cabinet not currently in use by the BMT lab.
- Researchers must have at least 2 identifiers on products and samples brought into the BMT lab- Product label, patient label containing MRN/CSN numbers, and an alphanumeric number. BMT lab do not provide labels.
- Any project study or IRB protocols will not be under our current BMT FDA registrations. If an FDA registration is needed, consult with BMT program and BMT lab directors. FDA registration should not be stated on the protocol without approval from the BMT program
- Any equipment that is needed from BMT Processing Lab will require training and documentation of training including decontamination procedures of the equipment. (Biologic safety cabinets, centrifuge, incubators, and controlled rate freezers).
- Prevention of Cross-Contamination Guideline: Research product will not be processed during BMT lab processing of patient products to avoid contamination. Research products should not be stored in any refrigerator used for storage of BMT patient products/samples.
- Equipment training checklist will include:

- Preparation of equipment: decontamination procedures of the equipment before use, settings, specimen prep, use of tools, check for cross contamination
- Operation of equipment: procedure
- Post operation of equipment: removal of specimen, check for contamination, decontamination procedures of the equipment after use.
- Work counter space
- Research specimens and documentation will be isolated to one workstation next to biologic safety cabinet assigned.
- BMT operations will take priority over any research operations. Researchers need to schedule their use of BMT lab space with the BMT lab well in advance before the date of use. If there is a conflict BMT lab operations takes priority.
- Any changes to the IRB protocol must be reviewed by all sections of the BMT program.
- Any violations in the agreement for the use of BMT lab space, equipment, cross-contamination, etc will be immediately brought to the attention of the BMT Laboratory Processing Medical Director.

Appendix H – Interferon Production

DATA COLLECTION FORM

ORIS PID: _____ **Date Completed:** ____ / ____ / ____

PI: Pierre Triozzi, M.D.

Study Number: CCCWFU 03716

Interferon Production				
Sample ID	Date	Stimulator (**#1**: anti-CD3 plus anti-CD28 antibody) or (**#2**: NY-ESO, survivin, and telomerase peptide mixtures)	IFN production	Notes

Appendix I – Frequency Counts from Flow Cytometry

DATA COLLECTION FORM

ORIS PID: _____ **Date Completed:** ____ / ____ / ____

PI: Pierre Triozzi, M.D.

Study Number:_____

Cell Frequency Counts				
Sample ID	Date	Cell type	Count/Frequency	Notes
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Appendix J – Neutrophil/Lymphocyte Data Form

DATA COLLECTION FORM

ORIS PID: _____ **Date Completed:** ____ / ____ / ____

PI: Pierre Triozzi, M.D.

Study Number: CCCWFU 03716

Neutrophil/Lymphocyte Data Collection Form				
Sample ID	Date	Neutrophil	Lymphocytes	Ratio